

# CROP ECOLOGY, PRODUCTION & MANAGEMENT

## Carbon Dioxide and Temperature Effects on Forage Dry Matter Production

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### ABSTRACT

Atmospheric CO<sub>2</sub> and temperature may significantly modify plant production. Grasslands occupy in excess of 25% of the Earth's land area, but grassland species have received limited attention from researchers studying climate change. A 3-yr study was conducted to determine the effects of elevated atmospheric CO<sub>2</sub> and temperature on dry matter (DM) harvested from the C<sub>3</sub> legume 'Florigrade' rhizoma peanut (RP, *Arachis glabrata* Benth.) and the C<sub>4</sub> grass 'Pensacola' bahiagrass (BG, *Paspalum notatum* Flüge). Both species were field grown in Millhopper fine sand (loamy siliceous Grossarenic Paleudult) in temperature-gradient greenhouses under different CO<sub>2</sub> (360 and 700  $\mu\text{mol mol}^{-1}$ ) and temperature conditions (baseline [B], B+1.5, B+3.0, and B+4.5°C, where B equaled ambient temperature). Plots (2 by 5 m) were harvested three times in 1996 and four times each in 1997 and 1998. Analyzed across years, yield increased 25% for RP ( $P = 0.02$ ) and tended to increase for BG (15%;  $P = 0.18$ ) with the near doubling of CO<sub>2</sub>, but there was species by CO<sub>2</sub> interaction ( $P = 0.06$ ) as a result of the greater response to CO<sub>2</sub> by the C<sub>3</sub> legume. There was a positive effect of increasing temperature on yield of both species. Averaged across species, yield increased 11% in 1996, 12% in 1997, and 26% in 1998 as temperature increased from B to B+4.5°C. Under well-watered conditions in this experiment, elevated CO<sub>2</sub> increased DM harvested of a C<sub>3</sub> legume and tended to increase that of a C<sub>4</sub> grass, while the yield response to increasing temperature was positive for both species.

ATMOSPHERIC CO<sub>2</sub> has steadily increased from preindustrial concentrations of 280  $\mu\text{mol mol}^{-1}$  and is predicted to double by the end of the 21st century (Barnola et al., 1994; Keeling and Whorf, 1994). Increasing atmospheric temperature is expected to accompany the increase in CO<sub>2</sub> (Jouzel et al., 1994). Jones (1997) suggested that DM production of C<sub>3</sub> grassland plants increases from 0.10 to 0.12% per 1  $\mu\text{mol mol}^{-1}$  increase in atmospheric CO<sub>2</sub>, while increases for C<sub>4</sub> plants are expected to be less. A review of effects of doubled CO<sub>2</sub> on forage plants suggests that DM production is likely to increase 30 and 10% for C<sub>3</sub> and C<sub>4</sub> species, respectively (Newton, 1991). Plants of the C<sub>4</sub> pathway have a built-in mechanism in the bundle sheath cells which allows them to maintain a higher CO<sub>2</sub> concentration around ribulose-1,5 biphosphate carboxylase/oxygenase (RuBisCO), enabling it to fix more CO<sub>2</sub> at ambient levels than does a C<sub>3</sub> plant. Therefore, an enriched CO<sub>2</sub> atmo-

sphere represents less change in CO<sub>2</sub> concentrations at the RuBisCO binding sites for C<sub>4</sub>, resulting in a smaller response for the C<sub>4</sub> compared with the C<sub>3</sub> plant. High temperature also generally increases accumulation of plant DM (Deinum and Dirven, 1974), but the literature on forages in this regard, and particularly tropical forages, is not abundant (Newton, 1991).

Dry matter production of grassland species determines herbivore carrying capacity and thus is a key factor affecting productivity of forage-livestock systems. Additionally, in many subtropical and tropical grasslands, large fluctuation in quantity of forage produced from season to season is a critical limitation to intake and livestock performance (Hardy et al., 1997), resulting in up to a 6-yr period to produce a slaughter animal on pasture (Mott and Moore, 1977). Consequently, understanding the impact of increasing CO<sub>2</sub> and temperature on productivity and seasonal distribution of DM of subtropical and tropical grassland species is particularly important. McGinn and Wedin (1997) propose the need for greater emphasis on these species when they indicate that most research on grassland responses to elevated CO<sub>2</sub> and climate change has occurred in temperate regions. The objectives of this experiment were to quantify effects of elevated atmospheric CO<sub>2</sub> concentration and temperature on (i) DM production of two perennating tropical-subtropical forage species, a C<sub>3</sub> legume (rhizoma peanut) and a C<sub>4</sub> grass (bahiagrass) and (ii) the seasonal distribution of that production.

### MATERIALS AND METHODS

#### Site and Species Description

The study was conducted in temperature-gradient greenhouses (TGGs) constructed over undisturbed field soil during 1996, 1997, and 1998 at the Irrigation Research and Education Park, University of Florida, Gainesville (29°38' N and 82°22' W). The average annual temperature is 21°C (average of years 1961–1990), with an average annual rainfall of 1342 mm. The soil was a Millhopper fine sand with a pH of 6.2 to 6.7. Soil organic matter at the site ranged from 12 to 15 g kg<sup>-1</sup>. For the analysis of soil P, K, Ca and Mg, the Mehlich-I extractant was used. Soil Ca concentration ranged from 240 to 340 mg kg<sup>-1</sup>, Mg from 98 to 115 mg kg<sup>-1</sup>, P from 79 to 97 mg kg<sup>-1</sup>, and K from 6 to 11 mg kg<sup>-1</sup>.

Stands of RP and BG were planted on 10 Apr. 1995 (Fritsch et al., 1999). Rhizoma peanut and BG are native to South America and are persistent perennials in North Florida. They initiate spring growth in March in most years and are produc-

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**Abbreviations:** B, baseline ambient temperature; BG, bahiagrass; DM, dry matter; PAR, photosynthetically active radiation; RP, rhizoma peanut; RuBisCO, ribulose-1,5-bisphosphate carboxylase/oxygenase; TGG, temperature gradient greenhouse.

tive until September or October when shorter days, cooler temperatures, and in some years drought stress limit growth.

### Treatments, Experimental Design, and Experimental Units

Treatments were the complete factorial arrangement of two levels of CO<sub>2</sub> concentration (360 and 700  $\mu\text{mol mol}^{-1}$ ), four temperatures (baseline [1.5°C above outside ambient], and

1.5, 3.0, and 4.5°C above baseline), and two forage species (Pensacola BG and Florigrade RP). The design was a split-strip-strip plot; CO<sub>2</sub> was the whole-plot factor and was allocated to greenhouses at random, with two greenhouses per level of CO<sub>2</sub>. Temperature and species were assigned in strips within each greenhouse (Fig. 1). The two strips for species (BG and RP) ran end to end, and species were allocated at random to the east or west side of a given greenhouse. Strips for temperature ran from side to side, and direction of the temperature gradient was assigned at random within a greenhouse. Plot size was 5 by 2 m for CO<sub>2</sub> × species × temperature treatment combinations.

The design of the TGGs was described in detail by Sinclair et al. (1995) and Fritsch et al. (1999). Each TGG was a free-standing unit, consisting of a semicircular arch structure made of galvanized steel and covered with SIXLIGHT (Taiyo Kogyo Co., Tokyo, Japan), a transparent polyethylene telephthalate film with 90% light transmission. The TGGs were 27.4 m long, 4.3 m wide, and 2.2 m high at the center, and were parallel to each other in north-south orientation.

### Fertilization and Irrigation

Fertilizer applied totaled 70 kg N, 30 kg P, and 58 kg K ha<sup>-1</sup> in 1996; 80 kg N, 36 kg P, 173 kg K, 65 kg Mg, and 128 kg S in 1997; and 80 kg N, 36 kg P, 152 kg K, 52 kg Mg, and 103 kg S in 1998. In 1996, fertilizer was applied in six equal splits (5 kg N ha<sup>-1</sup> equivalent) during each growth period preceding Harvests 2 and 3. Prior to Harvest 1, 10 kg N, 4 kg P, and 8 kg K were applied. In 1997 and 1998, 5 kg N ha<sup>-1</sup> was applied at four dates per growth period (total of 20 kg N ha<sup>-1</sup> per period) during each of four growth periods for a total of 80 kg N ha<sup>-1</sup> yr<sup>-1</sup>. The N source was ammonium nitrate and the S source was magnesium sulfate.

The irrigation system had 26 spaced microjet spray heads along each side of a greenhouse. During the 8- to 9-mo growing period, irrigation was applied three times per week with a double-overlapping microjet sprinkler system. This method of application provided 7 to 8 mm d<sup>-1</sup> on average during this period. During the winter months the rate was reduced approximately in half. The total annual irrigation averaged for the 3 yr, and expressed as a rainfall equivalent, was 2020 mm yr<sup>-1</sup>.

### Weed and Pest Control

Plots were hand weeded. During winter the major weed was "narrow leaf cudweed" (*Gnaphalium falcatum* Lam) of the Compositae family; during summer it was longstalked Phyllanthus (*Phyllanthus tenellus* Roxb.) of the Euphorbiaceae family, a C<sub>4</sub> plant. Phyllanthus was very prolific at the warmest temperatures and enriched CO<sub>2</sub>. Weeds were more prevalent in RP than in BG.

The major pests of RP were spider mites (*Tetranychus urticae* Koch), black aphids (*Aphid* spp.), and thrips, very likely the species *Frankliniella fusca* (Hinds), *Thripidae*. Mole crickets (*Scapteriscus borelli*) were the only pest on BG. Labeled sprays and baits were applied on an as-needed basis for control.

### Temperature Control

The methodology to control temperature and CO<sub>2</sub> was based on the TGG infrastructure used by Okada et al. (1995) with modified hardware as described by Sinclair et al. (1995). To obtain the desired temperature during daytime, solar radiation reaching the cells served as the primary energy source, and additional heated air (120°C) was infused into the chamber

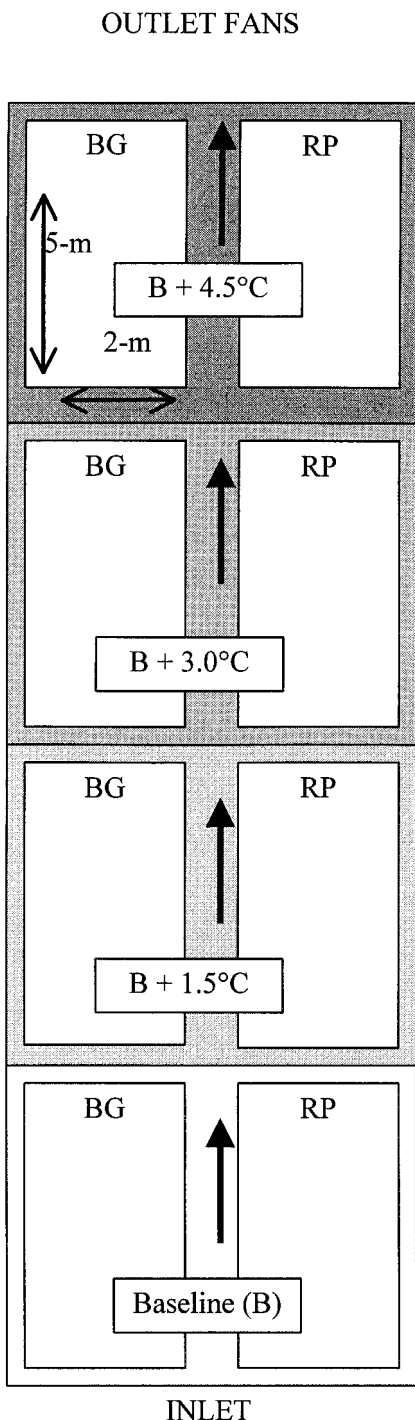


Fig. 1. Example of treatment layout in a temperature-gradient greenhouse (not to scale). Unidirectional arrows indicate the direction of air flow. One greenhouse represents one replicate of one CO<sub>2</sub> treatment.

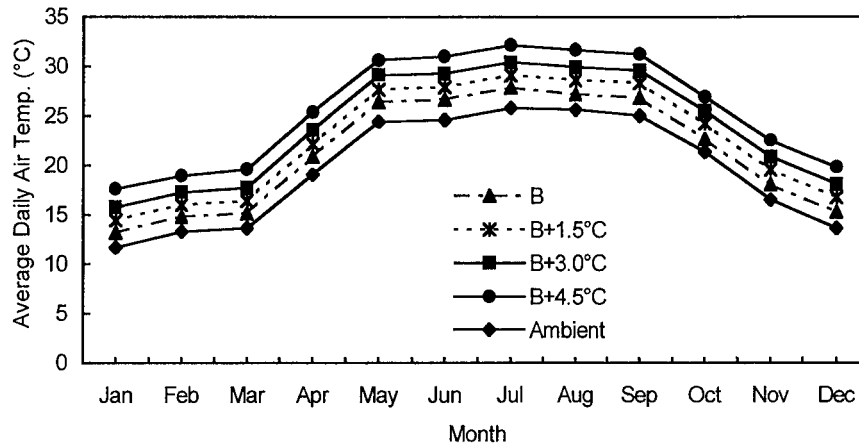


Fig. 2. Monthly averages of average daily air temperature in Greenhouse 1 for baseline (B) through B+4.5°C temperature treatments during 1996.

at 5.5-m increments along its length. Heat was added at night and under low solar radiation conditions when natural solar radiation was insufficient to maintain the desired temperature. Horton (1999) calibrated the ventilation fans and described the air flow and temperature patterns in the greenhouses under various conditions. By injecting heated air at 5.5-m increments and by placing overhead ceiling fans at each 5.5-m increment, a stepwise gradient of 1.5°C was created. An exhaust fan was used to regulate the temperature gradient. This exhaust fan was mounted at the warm end of the greenhouse and had computer-regulated variable speed control, which was manipulated to reduce the deviation from the desired temperature gradient. When the target temperature difference (4.5°C) between the two extreme plots inside the TGG was exceeded, the exhaust fan speed was increased. When the temperature difference was below the target difference, the exhaust fan speed was decreased. The operation of the exhaust fan was controlled by a controller/data logger system based on aerial thermocouples that monitored air temperature at 0.9 m above soil level over the B and B+4.5°C plots in each greenhouse.

The desired temperature gradient in each TGG was maintained (Fig. 2; 1996 data). Soil temperature was also monitored with thermocouples at a 0.09-m soil depth, below the aerial thermocouples. This response is shown for one greenhouse during the 1996 season (Fig. 3).

### Data Collection and Processing

The chambers were controlled by a Keithley Metrabyte data logging system (Keithley Instruments, Boston, MA) operated by FIX DMACS software (Intellution, Norwood, MA). Data were also collected by this controller/data logger system. Information from temperature sensors in the chambers was collected every minute.

In consideration of the dynamic temperature variation, moving averages were recorded at 20-min intervals over 24-h periods. Each 24-h segment was stored as a separate binary file on a computer hard drive. Minimum, maximum, and mean values were obtained for each day, and from daily values, monthly summaries were obtained. An example summary of these temperature records collected for the baseline cell in one greenhouse is presented (Fig. 4). Photosynthetically active radiation (PAR) was measured with a calibrated quantum sensor (Model LI-6200, LI-COR, Lincoln, NE) located outside of the TGGs.

### CO<sub>2</sub> Control

Carbon dioxide was provided from a supply tank outside the TGGs and injected into a predilution distribution system placed approximately 1.2 m inside the inlet end of the TGG. The CO<sub>2</sub> concentration was measured every 20 s at 0.6 m above soil surface and about 6-m downflow from the injection

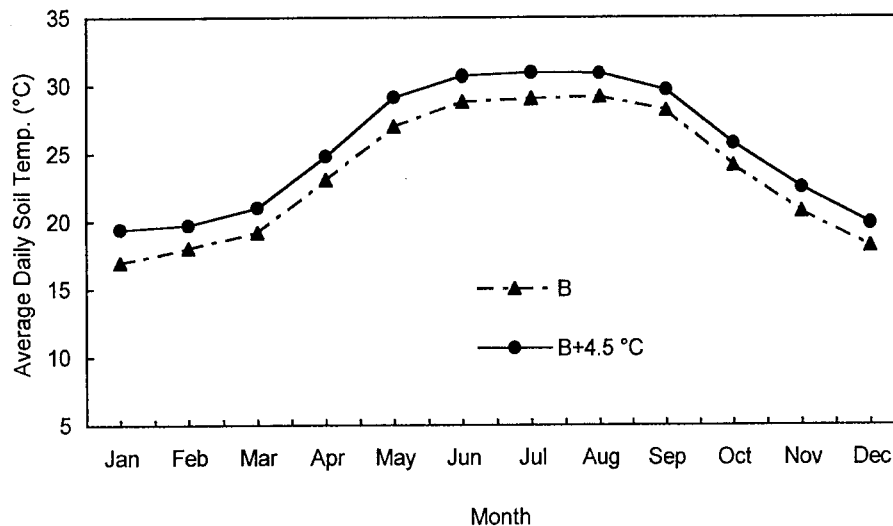


Fig. 3. Monthly averages of average daily soil temperature in Greenhouse 1 for baseline (B) and B+4.5°C temperature treatments during 1996.

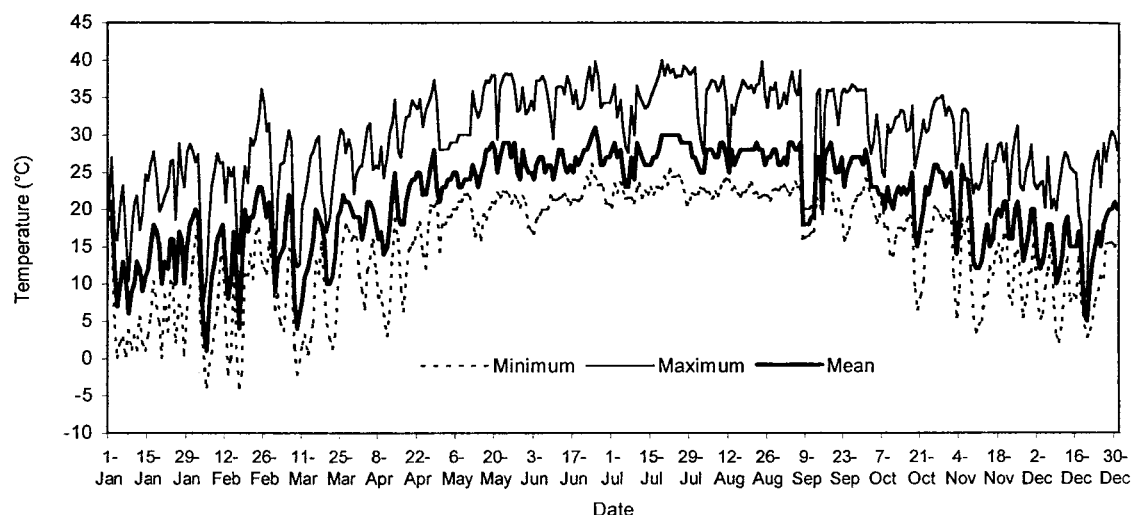


Fig. 4. Daily mean, minimum, and maximum air temperature for baseline temperature in Greenhouse 1 during 1996. These values were extracted from measurements over each 24-h period at 20-min intervals.

distribution system. The volume of  $\text{CO}_2$  released to maintain  $700 \mu\text{mol mol}^{-1}$  was changed with a variable gas valve depending on the speed of the exhaust fan using a proportional/integral adjustment algorithm. Liu (1999) conducted several performance tests of the  $\text{CO}_2$  injection and control system. She found that the  $\text{CO}_2$  controller maintained the long-term average  $\text{CO}_2$  concentration to within approximately  $5 \mu\text{mol mol}^{-1}$  of the  $700 \mu\text{mol mol}^{-1}$  setpoint concentration. Within each 2.5-h-long test by Liu (1999), the standard deviation of  $\text{CO}_2$  concentration ranged from 31 to  $53 \mu\text{mol mol}^{-1}$ . A very low rate of supplemental  $\text{CO}_2$  was continuously injected at the start of Cells 2, 3, and 4. As a result, no  $\text{CO}_2$  depletion gradients were observed along the TGG from Cells 1 to 4.

### Field Harvests

The harvest schedule chosen for 1996 was similar to the three-cut system recommended for RP hay in Florida. Rapid growth in the TGGs led to lodging of forage before harvests in 1996, therefore in 1997 and 1998 a four-cut system was used. Harvest dates were 14 June, 13 Aug., and 18 Nov. 1996; 9 May, 3 July, 21 Aug., and 21 Nov. 1997; and 18 May, 6 July, 8 Aug., and 9 Nov. 1998. Above-ground biomass of RP and BG was harvested with a sickle-bar mower to a 3-cm stubble. The fresh forage yield for each plot was weighed, and a subsample of approximately 800 g fresh weight was taken for DM determination. The subsample was oven dried for 48 h at  $60^\circ\text{C}$ . Distribution of DM was expressed within years as the

percentage of total annual production harvested at each cutting date.

### Statistical Analyses

Data were analyzed by mixed model methodology. In all models,  $\text{CO}_2$  concentration, temperature, species, and their interactions, were considered fixed effects. Greenhouses nested within  $\text{CO}_2$  levels were modeled as random effects. Data were analyzed by PROC MIXED (SAS Institute Inc., 1996). The nature of harvest (for seasonal distribution data) and temperature effects was assessed by orthogonal polynomial contrasts. All means reported in the text are least square means. Values of  $P \leq 0.10$  were considered significant.

## RESULTS AND DISCUSSION

### Temperature and Radiation

Mean temperatures were higher for all months in 1998 compared with 1996 (Table 1). Year 1998 (starting in fall of 1997) was considered an El Niño year, during which Florida experienced drier summers and warmer winters than usual (Green et al., 1997). June 1998 was exceptionally warm, with record high temperatures on 7 d.

Distribution of PAR throughout the year was similar during 1996 and 1997 (Fig. 5). In 1998, PAR increased by almost  $10 \text{ mol photon m}^{-2} \text{ d}^{-1}$  in June and by 5 mol

Table 1. Monthly ambient air temperatures (minimum, maximum, and mean) for 1996 to 1998.

Month	1996			1997			1998		
	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean
°C									
Jan	3.9	19.2	11.6	7.1	20.5	13.8	8.1	20.8	14.4
Feb	5.4	21.4	13.4	9.7	23.2	16.4	7.6	20.2	13.9
Mar	6.4	21.0	13.7	14.2	27.4	20.8	8.0	22.4	15.2
Apr	10.3	26.8	18.5	12.0	25.9	18.9	12.8	27.6	20.2
May	16.9	31.8	24.3	16.4	29.7	23.1	17.4	32.7	25.1
Jun	18.9	32.0	25.4	19.7	30.7	25.2	21.4	36.2	28.8
Jul	20.5	33.3	26.9	21.3	33.2	27.2	23.0	34.5	28.7
Aug	20.1	32.2	26.2	20.7	33.3	26.9	21.9	33.7	27.8
Sep	18.4	31.7	25.1	19.0	33.3	26.2	21.8	30.8	26.3
Oct	14.4	27.4	20.9	14.4	27.9	21.2	15.8	29.7	22.8
Nov	10.2	23.6	16.9	9.2	22.6	15.9	13.6	26.8	20.2
Dec	6.9	20.5	13.7	7.6	18.7	13.2	9.2	23.9	16.6



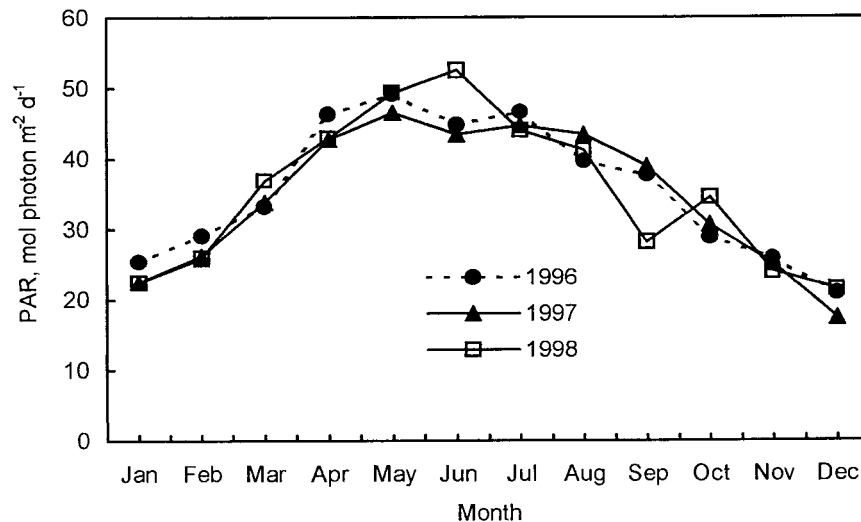


Fig. 5. Photosynthetically active radiation (PAR) for the 1996–1998 seasons. Values for each month are daily averages.

photon  $\text{m}^{-2} \text{d}^{-1}$  in October, but decreased by almost 10 mol photon  $\text{m}^{-2} \text{d}^{-1}$  in September, compared with 1996 and 1997. These differentials in PAR are presumably due to differences in cloudiness associated with presence or absence of rainfall.

### Treatment Effects on Total DM Harvested

Probability values are presented in Table 2 for the effects of year, species,  $\text{CO}_2$ , temperature, and their interactions on DM harvested. Analysis across years revealed that total DM harvested was affected by year, species, and a species  $\times$  year interaction ( $P < 0.01$ ). Also across years,  $\text{CO}_2$  and temperature affected the response ( $P = 0.04$  and  $P = 0.01$ , respectively) and there was a significant  $\text{CO}_2 \times$  species interaction ( $P = 0.06$ ). Analysis of variance subsequently was run within years because of species  $\times$  year interaction ( $P < 0.01$ ) and because RP stands, which were planted in 1995, usually take 2 to 3 yr for complete stand establishment (Valentim et al., 1987; Rice et al., 1995).

### $\text{CO}_2 \times$ Species Interaction Effect on Total DM Harvested

There was a  $\text{CO}_2 \times$  species interaction across years (Table 3). Total DM harvested was greater for RP than

BG under ambient and enriched  $\text{CO}_2$  conditions ( $P < 0.01$ ). The interaction occurred because there was only a trend for BG total DM harvested to be greater at elevated  $\text{CO}_2$  concentrations while RP yield increased 25% from 360 to 700  $\mu\text{mol CO}_2 \text{ mol}^{-1}$ . Results from 3 yr suggest that both species may benefit from  $\text{CO}_2$  enrichment, but RP clearly benefited to a greater extent than BG. These results are consistent with DM yield responses attributed to the “fertilization effect” of increased  $\text{CO}_2$  on  $\text{C}_3$  plants like RP and  $\text{C}_4$  species like BG (Kimball, 1983; Newton, 1991). Greater proportional response of RP than BG at elevated  $\text{CO}_2$  concentration occurs because at ambient  $\text{CO}_2$ ,  $\text{C}_4$  species have a mechanism which allows them to maintain higher  $\text{CO}_2$  concentration around Rubisco than is the case for  $\text{C}_3$  species.

When analyzed within year (Table 2), the  $\text{CO}_2 \times$  species interaction was significant only in the third year ( $P = 0.02$ ), although trends were present in 1996 ( $P = 0.22$ ) and 1997 ( $P = 0.18$ ). Decreasing standard errors for DM harvested from 1996 to 1998 likely reflect greater uniformity of more fully established stands in later years and allowed for a stronger test of the interaction. Total DM harvested was greater under enriched  $\text{CO}_2$  conditions in all years for RP, but only in 1997 there was a strong trend ( $P = 0.17$ ) for a  $\text{CO}_2$  effect on BG (Table 4). Near doubling of ambient  $\text{CO}_2$  concentration increased total DM harvested from RP by 27% in 1996, 28% in 1997, and 23% in 1998.

### Species $\times$ Year Interaction Effect on DM Harvested

The species  $\times$  year interaction occurred because RP DM harvested increased each year from 1996 through

Table 2. Levels of probability ( $P$  value) for the effect of year (Yr),  $\text{CO}_2$ , species (Sp), temperature (Temp), and their interactions on total dry matter (DM) harvested.

Source of variation	1996–1998	1996	1997	1998
Yr	<0.01			
$\text{CO}_2$	0.04	0.14	0.01	<0.01
$\text{CO}_2 \times \text{Yr}$	0.77			
Sp	<0.01	0.01	<0.01	<0.01
$\text{Sp} \times \text{Yr}$	<0.01			
$\text{Sp} \times \text{CO}_2$	0.06	0.22	0.18	0.02
$\text{Sp} \times \text{CO}_2 \times \text{Yr}$	0.46			
Temp	0.01	0.30	0.23	<0.01
$\text{Temp} \times \text{Yr}$	0.18			
$\text{Temp} \times \text{CO}_2$	0.39	0.54	0.59	0.84
$\text{Temp} \times \text{CO}_2 \times \text{Yr}$	0.97			
$\text{Sp} \times \text{Temp}$	0.19	0.82	0.35	0.17
$\text{Sp} \times \text{Temp} \times \text{Yr}$	0.93			
$\text{Sp} \times \text{Temp} \times \text{CO}_2$	0.50	0.18	0.36	0.82
$\text{Sp} \times \text{Temp} \times \text{CO}_2 \times \text{Yr}$	0.53			

Table 3. Interaction effect of species and atmospheric  $\text{CO}_2$  concentration on total annual dry matter (DM) harvested averaged across years (1996–1998).

$\text{CO}_2$	Bahiagrass	Rhizoma peanut	$P$ -value	SE
$\mu\text{mol mol}^{-1}$	Mg $\text{ha}^{-1}$			Mg $\text{ha}^{-1}$
360	7.8	15.5	<0.01	0.50
700	9.0	19.3	<0.01	0.50
$P$ -value	0.18	0.02		

**Table 4. Interaction effect of atmospheric CO<sub>2</sub> concentration and species on total annual dry matter (DM) harvested during 1996–1998.**

Year	Bahia grass			Rhizoma peanut			SE
	CO <sub>2</sub> (μmol mol <sup>-1</sup> )		P-value	CO <sub>2</sub> (μmol mol <sup>-1</sup> )		P-value	
	360	700		360	700		
	Mg ha <sup>-1</sup>			Mg ha <sup>-1</sup>			Mg ha <sup>-1</sup>
1996	8.8	10.2	0.31	13.2	16.8	0.06	0.86
1997	7.4	9.1	0.02	14.6	18.7	0.01	0.75
1998	7.1	7.8	0.31	18.5	22.8	<0.01	0.45

1998, while that of BG decreased each year (Table 5). For RP this response is thought to be associated with more fully established stands and specifically in 1998 with higher temperatures (Table 1) and higher PAR due to reduced cloud cover and dry weather during June and to a lesser extent during October 1998 (Fig. 5). Averaged across CO<sub>2</sub> levels, DM harvested from BG was 9.5, 8.3, and 7.5 Mg DM ha<sup>-1</sup> yr<sup>-1</sup> in 1996, 1997, and 1998, respectively, constituting decreases of 13% from 1996 to 1997, and 10% from 1997 to 1998. The lower yields of BG as years progressed were probably associated with N deficiency. The fertilization rate of 70 to 80 kg N ha<sup>-1</sup> yr<sup>-1</sup> was relatively low for infertile sandy soils under well-watered conditions and with removal of plant biomass by clipping. If N was not limiting, DM yield of BG would be expected to equal or exceed that of RP. Yield of BG receiving 100 kg N ha<sup>-1</sup> yr<sup>-1</sup> growing under normal field conditions is often higher than yield of RP, and N concentration of bahiagrass typically ranges from 12 to 20 g kg<sup>-1</sup> (Dunavin, 1992; Williams, 1994). Under the conditions of the current study, RP outyielded BG (averaged across CO<sub>2</sub> levels) by 58% in 1996 and by more than 100% in 1997 and 1998. Rhizoma peanut was not N limited because it was well nodulated with apparently effective nodules. Leaf N for BG (averaged across CO<sub>2</sub> and temperature levels) for 1996 and 1997 was 9.0 and 8.5 g kg<sup>-1</sup>, respectively (Newton, 1999). A similar situation of declining grass yield over time was reported by Newton (1991) for perennial ryegrass (*Lolium perenne* L.), which exhibited a limited response to CO<sub>2</sub> when grown at low levels of N. In the current study, it is not known if low N concentration simply limited BG yield at both CO<sub>2</sub> concentrations, or if low N limited the ability of BG to respond to elevated CO<sub>2</sub>. Newman et al. (1999) reported no CO<sub>2</sub> × N rate interaction for DM harvested when BG was fertilized at 80 (as in current study) and 320 kg N ha<sup>-1</sup>, supporting the idea that N concentration was not limiting the yield response of BG to CO<sub>2</sub>. Nevertheless, the trend toward a CO<sub>2</sub> effect on BG production

(across years) was comparable to those typically reported for C<sub>4</sub> species (Newton, 1991).

### Temperature Effects on DM Yield

When analyzed across years there was a linear effect of temperature ( $P = 0.01$ ) on DM harvested. There were no interactions involving temperature, although there was a trend ( $P = 0.18$ ) toward a temperature × year interaction (Table 2). Within years, total DM yield increased linearly with increasing temperature in 1996 and 1997 ( $P < 0.10$ ), but in 1998 there were both linear ( $P < 0.01$ ) and quadratic ( $P = 0.05$ ) effects (Table 6). From baseline to B+4.5°C, yield increased 11% in 1996, 12% in 1997, and 26% in 1998.

### Seasonal Distribution of DM Harvested

The distribution of DM harvested expressed by harvest as a percentage of total annual DM harvested, was not affected by CO<sub>2</sub>, temperature, or their interactions, but was different for the two species and was affected by a species × year × harvest number interaction (Fig. 6). Analyses by year showed a species × harvest number interaction each year; therefore, results are reported within species and year. Proportion of total DM harvested for RP decreased from first to last harvest in all years. A similar response was observed for BG only in the first year when harvested three times, but when BG was harvested four times the proportion of total DM increased from first to last harvest. In 1997 and 1998, first-harvest BG yields contributed approximately 15% of the total-season DM compared with more than 25% for RP. These results agree with literature reports for seasonal distribution of DM yields of BG and RP (Williams, 1994). She found that for RP highest yields occurred in the spring and early summer, followed by a gradual linear decrease in DM production in fall. Lack of temperature effect and species × temperature interaction suggests that factors other than temperature

**Table 5. Interaction effect of species and year on total annual DM harvested averaged across atmospheric CO<sub>2</sub> concentrations and temperatures.**

Year	Bahia grass	Rhizoma peanut	P-value†	SE
	Mg ha <sup>-1</sup>			Mg ha <sup>-1</sup>
1996	9.5a‡	15.0a	<0.01	0.60
1997	8.3b	16.7b	<0.01	0.53
1998	7.5c	20.6c	<0.01	0.32

† P-value for species effect within year.

‡ Year means within a species are not different ( $P > 0.1$ ) if followed by the same letter.

**Table 6. Temperature effect on total annual DM harvested averaged across CO<sub>2</sub> concentrations and species.**

	Temperature (°C)				Polynomial contrast‡	SE
	B†	B+1.5	B+3.0	B+4.5		
	Mg ha <sup>-1</sup>					Mg ha <sup>-1</sup>
1996	11.7	12.1	12.1	13.0	L	0.60
1997	11.9	12.3	12.3	13.3	L	0.55
1998	12.1	14.1	14.8	15.2	L*, Q	0.41

† Baseline temperature, defined as ambient temperature in the greenhouse.

‡ Orthogonal polynomial contrast of temperature effect within year. L = linear, Q = quadratic; letter with no symbol ( $P \leq 0.1$ ); \*\* ( $P \leq 0.01$ ).

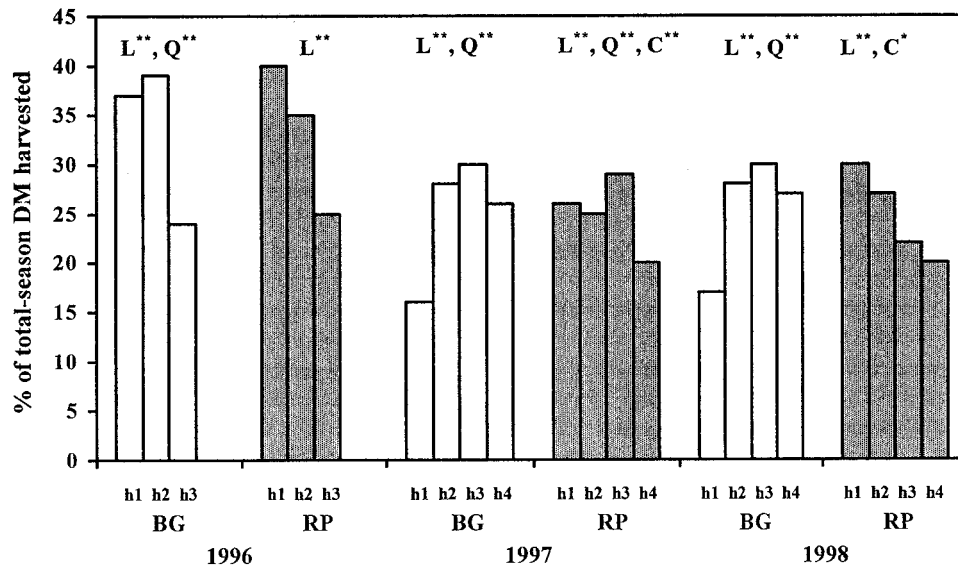


Fig. 6. Percentage of total-season DM harvested of bahiagrass (BG) and rhizoma peanut (RP) by harvest. There were three harvests (h) in 1996 and four in 1997 and 1998. L = linear, Q = quadratic, and C = cubic effect of harvest number. \*\*,  $P \leq 0.01$ ; \*,  $P \leq 0.05$ .

likely are influencing the seasonal patterns of growth of BG and RP.

## SUMMARY AND CONCLUSIONS

Grassland species, and in particular tropical and subtropical species, have received limited attention from researchers studying climate change (McGinn and Wedin, 1997). The objective of this study was to determine the effects of elevated atmospheric  $\text{CO}_2$  and temperature on DM harvested and seasonal distribution of RP and BG production.

Elevated  $\text{CO}_2$  increased DM harvested of RP by 25% compared with that at ambient  $\text{CO}_2$ , while for BG there was only a trend ( $P = 0.18$ ; 15%) toward increasing DM harvested at the higher  $\text{CO}_2$  level. These data are comparable to those in the literature for other  $\text{C}_3$  and  $\text{C}_4$  species, and these effects can be attributed primarily to differences in  $\text{C}_3$  and  $\text{C}_4$  carbon fixation pathways. Increasing temperature increased DM harvested of both species from 11 to 26% during the 3 yr, but did not affect seasonal distribution of DM. Averaged across  $\text{CO}_2$  and temperature levels, DM harvested of RP increased each year, likely in part because of more complete stand establishment in the second and third years and to increased PAR (June and October) and higher temperatures in the third year. In contrast, BG had lower DM harvested each succeeding year, in part because of a N limitation, evidenced by leaf tissue N concentrations that were below  $10 \text{ g kg}^{-1}$ .

These data suggest that yield responses of  $\text{C}_3$  forage legumes to increasing atmospheric  $\text{CO}_2$  concentration may be in the range of 20 to 30% in environments where soil water is not limiting growth. Yield responses of  $\text{C}_4$  grasses to  $\text{CO}_2$  are less pronounced and in this experiment were not statistically significant ( $P \leq 0.10$ ) in any year or across years. Higher growth temperatures increased DM harvested of both species. Thus, when soil water is not limiting, conditions of elevated  $\text{CO}_2$  and temperature (i) are likely to increase productivity of

these subtropical and tropical grassland species, (ii) may increase productivity of  $\text{C}_4$  species, but the response to  $\text{CO}_2$  is less than that of  $\text{C}_3$  species if it occurs at all, and (iii) may have little effect on seasonal distribution of DM.

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## Control of Interplot Interference in Grain Maize: A Multi-Site Comparison

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### ABSTRACT

Interplot interference is known to be potentially a major source of bias in cultivar trials of several plant species, but there are few published results concerning grain maize (*Zea mays* L.) in France. Two series of field experiments were conducted in the North and in the South of France from 1994 to 1996 to quantify interference in grain maize trials and compare methods of control. Each experiment typically consisted of a reference trial with two or three unharvested rows on each side of each plot, a four-row-plot trial with one unharvested row on each side of each plot, and a two-row-plot trial. Trials had seven cultivars in 1994 and 1996, and four additional cultivars in 1995. Interference was found to occur in two-row-plot trials and to be related to plant height. When a cultivar was 10 cm shorter than each of its neighbors, its yield was reduced by 0.16 Mg ha<sup>-1</sup> in the North series and by 0.30 Mg ha<sup>-1</sup> in the South series on average. Interference appeared much lower in the four-row-plot trials. Methods for controlling interference were assessed by comparing their cultivar estimates with those from the reference trials. Bias due to interference in the two-row-plot trials was reduced by using models for interference, but the four-row-plot trials appeared as a more reliable method to avoid such bias.

INTERFERENCE BETWEEN PLOTS is known to occur in cultivar trials for many cultivated species (see Talbot et al., 1995; Kempton, 1997 and references therein). It is mainly caused by competition between the cultivars

located on neighboring plots, and it may artificially favor some cultivars and penalize others. Interplot interference has been extensively studied for cereals, in particular for wheat (*Triticum aestivum* L.), where a relationship with height has been recognized (Kempton et al., 1986; Goldringer et al., 1994; Clarke et al., 1998). In grain maize cultivar trials, interference is potentially a major concern, since plots usually consist of only two to four seeded rows and cultivars may exhibit large phenotypic differences. Consequently, there may be large interference effects exerted on a large proportion of each plot. Interference has been found to occur for grain maize in a few studies in the USA (Kiesselbach, 1923; Genter, 1958; Pendleton and Seif, 1962; Ziegler, 1980; Esgar and Bullock, 1999). Interference effects may be due to differences in plant height, maturity, vigor, leaf area, density, or planting date. On the contrary, Olson (1928) and Bowman (1989) found little evidence of interference. Fewer published references exist for grain maize in Europe, but interference effects have been reported in France (Lorgeou, 1986) and in Belgium (Van Waes, 1997).

Interplot interference is a source of bias for cultivar comparisons and so needs to be controlled as well as possible. There exist several alternative or complementary methods to do this, either when defining plot size, choosing the design, or analyzing the data (Kempton, 1997; Monod et al., 1997).

First, interference may be limited by enlarging plot size and leaving border rows of each plot unharvested;

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